RESEARCH ARTICLE

Phytoremediation Potential of *Glycine max L* and Empty Palm Fruit Bunch Ash (Potash) in soils co-contaminated with Chromium and Anthracene

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ABSTRACT: Phytoremediation is an assuring technology for the remediation of sites polluted with inorganic and organic contaminants. The present study assessed the remediation potential of *Glycine max L* in a co-contaminated soil with Chromium and anthracene and the effects of potash applied as amendments. Soils were contaminated with potassium chromate salt and anthracene at one level (100mg/kg of Cr+100/mg/kg of anthracene) and arrayed in completely randomized design with 3 replicates. *Glycine max L* seeds were planted. Potash amendments were applied at 30g/kg as solution to each soil surface at doses of 10g/kg for three weeks. There was 49% inhibition of shoot dry matter of *Glycine max L* with respect to control treatments. Application of potash to anthracene and Cr-anthracene co-contaminated soil decreased the residual anthracene to 29±1.17 f and 22±1.16 de respectively. This result proposed that *Glycine max L* is an assured candidate for uptake of Cr and dissipation of anthracene in co-contaminated soils amended with potash.

KEYWORDS: Phytoremediation, Empty palm fruit bunch ash, Co-contaminated soil

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INTRODUCTION

Rapid industrial development and exceptional agricultural activities have led to a large amount of organic compounds and heavy metals entering the environment, leading to serious co-contamination [1, 2].The coexistence of organic and inorganic pollutants has attracted increasing attention worldwide, as the two different types of pollutants often interact with each other, leading to more complicated circumstances and intensifying environmental remediation tasks [2]. Several studies have demonstrated that the combination of these two types of contaminants could potentiate great environmental risk to vegetation, soil microbial and human health. Phytoremediation, also called eco-friendly remediation, green rernediation, botano-remediation, agro remediation, or vegetative remediation is considered a publicly appealing remediation technology that uses vegetation and associated micro biota, soil amendments and agronomic techniques to remove, contain, or render the heavy metals harmless in the soil [3,4,5]. Plant-based technologies can target both inorganic and organic pollutants. There is an increasing interest to study the phytoremediation of co-contaminated soils [6-9]. This study will principally focus on phytoremediation potential of *Glycine max L* on soil co contaminated with anthracene and chromium using potash (empty palm fruit bunch) as soil amendments. Potash is a carbonaceous product produced from empty palm fruit bunch. It has many potential benefits on soil properties as an increase in soil biological activities, increase in pH and nutrient uptake. It also possesses anions like phosphate, chloride, nitrate and sulphate which acts as a degrading agent [10-13]. The present study was conducted to assess the effects of potash application as soil amendment on the degradation of anthracene and the concurrent uptake of chromium by *GlycinemaxL* in co-contaminated soil. Anthracene was used as an example of organic compounds in this research because anthracene present a class of organic compounds that exist mostly in polluted sites with hazardous material contamination while Cr is among the ten prime concern contaminants. *Glycine max L* was used as a result of its advantageous features like N-fixing

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bacteria, short life cycle, broad tap root system and handling easiness [14].

2.0 MATERIALS AND METHODS

Dry seeds of *Glycine max L* were obtained from National Seeds Service, Umudike. Potassium chromate salts was acquired from Finlab, Nigeria. Soils (0-20cm) deepness with moisture content 1.29%, CEC 640 Cmol/kg, pH 4.8., organic matter 1.97%, total nitrogen content 0.1 5%, silt 3.46% sand 91%,and clay 5.14% (sandy loam) were heap upusing soil auger from 3 specified sampling points from a farm land in Federal University of Technology Owerri, Imo state which lies within latitudes 5°27□ 50.23''North and longitude 7°02□ 49.33'' East at height above sea level of 55m.

2.1 Soil preparation / Soil spiking

The compounded soil amassed was subtly sifted to-pass through a 2mm sieve to take out foreign and bristly particles, air-dried and filled into a total of 2l pots for spiking. Plastic pots of 11 cm high were used for the research. Pots were filled with spiked soil weighing 1kg. Soil was spiked with anthracene by dissolving 100 mg of anthracene in 20 ml of acetone. 100 mg /kg of Cr were made ready for use by dissolving 0.597 g of $K_2C_{r2}O_7$ and added solely in anthracene spiked soils. The spiked soil was thoroughly mixed by sieving and kept in a dark room for equilibration for 8 days before planting.

2.2 Planting

The experiment was arranged in a completely randomized design with 7 treatments and three replicates. Pots spiked with anthracene and Cr were planted with *Glycine max* seeds, pots without *Glycine max* plant were used as control to detect the non-plant facilitated dissipation of anthracene. Potash was added into the pots 5 weeks after planting respectively. This was spitted into three parts and applied each week for 3 weeks. Treatments comprises the control soil (without amendments) and 30 g kg^{-1} of potash were applied as solutions to each soil surface at doses of 10 g kg^{-} for three weeks to curtail the effect of the amendments on plant growth, and as indicated by [7], split applications were more effectual. Pots were watered when needed and the leachates from all pots were recovered using the tray and put back to the soil. Plant samples were taken from the pots 70 days after planting, washed and oven dried to stable weight at 70°C for 50 h. The dried samples were weighed to decide dry matter yield which was used for plant analysis. For vegetated pots, rhizosphere soil samples were collected. Samples were analyzed for Cr uptake by using Varian AA240 Atomic Absorption Spectrophotometer according to the method of [15] (American Public Health Association) and residual soil anthracene was carried out by using soxhlet extraction method [16].

2.3. Evaluation of Cr uptake and Anthracene dissipation

To assess the efficiency of *Glycinemax L* in Cr assimilation and in the removal of anthracene, the following criterion were determined: stover dry weight; .Metal concentration in plant tissues; Translocation factors (TFs) which is the transfer of metals from the roots to the aboveground parts was ascertained as the metal in shoots to the metal in roots ratio and was clearly defined by the formula; TF = A shoot/A root. Bioconcentration factors (BCFs) which measure the accumulation of metals in plants is ascertained as the ratio between metal concentration in plant tissues and total metal initial soil concentration. BCFs is conveyed by the formula; BCFshoots= Ashoot / Asoil. BCFroots = Aroot / Asoil. Where Ashoot and Aroot are metal concentrations in the shoot (mg kg') and root of plants (mg kg^{-1}), respectively, and Asoil is the metal concentration in the soil (mg kg^{-1}) [4]. Soil PAH residual concentration was calculated as the final soil residual anthracene concentration after G.C analysis. Anthracene dissipation (%) was calculated as; 100 x [Ci-Cs/Ci}. Where, Cs is the concentration of PAH in each treatment. Ci is the initial PAH concentration present in the soil [7].

2.4. Statistical analysis

All data composed were subjected to statistical analysis using Statistical Package for Social Science (SPSS version 21 software package) (SPSS lnc, Chicago, IL, USA). Treatment effects were evaluated by analysis of variance (ANOVA). When a significant difference was noticed between treatments, multiple comparisons were made by Tukey HSD test. Differences were considered significant at $p < 0.05$.

3.0. RESULTS AND DISCUSSION

3.1. Growth response

No detectable toxic symptoms were observed in the test crop–*Glycine max (*L) plant. Root and shoot dry matter weight of *Glycine max L* was affected by Cr and anthracene co-contamination. 100mg kg' of Cr really decreased the shoot dry matter of *Glycine max L* by 21% when related with control treatments [Table 1]. There was 49% reduction of shoot dry matter of *Glycine max* relative to control treatments when 100 mg Cr kg was mixed with 100 mg kg^{-1}anthracene. Compared to control treatments, the shoot dry weight of *Glycine max* increased significantly to 0.38±0.02g $gpot^{-1}$ with the addition of potash in single Cr contamination and increased to 0.34±0.02g pot^{-1} in co-contamination with Cr and anthracene. In anthracene contaminated soil; the shoot biomass was enhanced with the addition of potash when compared with shoot biomass with treatment and no amendment by 26% [Table. I].

Table 1: Plant dry matter yield as influenced by potash in the remediation of co-contaminated soil using *Glycine max*.

Treatments	Shoots Biomass (g/pot)	Root Biomass (g/pot)
Soil + G.max	0.33±0.01a	0.21±0.01a
100mg/kg Cr+G.max	0.26±0.01a	0.19±0.01a
100mg/kg Cr+G.max+p	0.38±0.01bc	0.27±0.01b
100PAH+G.max	0.28±0.01c	0.17±01bc
100PAH+G.max +p	0.38±0.01cd	0.22±0.01bc
100PAH+100mg/kg Cr+G.max	0.17±0.01c	0.18±0.01b
100PAH+100mg/kg Cr + G.max+p	0.34±0.02c	0.20±0.02b

3.2. Cr concentration in plants tissues as influenced by potash application

Seventy days after planting, (70DAP), the application of potash in the present research did not increase the concentration of Cr in shoot of *G.max* when soil was contaminated with Cr alone and Cr - anthracene. As shown in Table 2, potash increased the root Cr concentration of *G.max* when soil was contaminated with Cr alone but in Cr - anthracene co-contaminated soils, the application of potash lowered the root concentration of Cr to 5.7±0.52mg kg^{1} (Table 2).

Table 2: Cr accumulation as affected by potash in the remediation of co contaminated soil using *Glycine max*

Treatments	Shoot Conc.(Mg/Kg)	Root Conc.(Mg/kg)
Soil+G.max	0.001±0.00	0.0034±0.00
100mg/kg Cr+G.max	2.4±0.28a	9.8±0.52a
100mg/kgCr+ G.max +p	1.3±0.40c	15±0.52c
100PAH+100mg/kgCr+G.max	4.2±0.40d	7.2±0.52cd
100PAH+100mg/kgCr+G.max +p	1.5±0.40d	5.7±0.52e

Different Letters indicate significant difference (TukeyHSD,p<0.05)
n=3, p=potash, l00PAH=100mg/kg anthracene.

3.3 Effects of potash on the translocation and bio concentration factors of Cr

The translocation factor (TF) value of Cr decreased with the application of potash in. single Cr contaminated soil and co-contaminated soil to 0.09±0.04c and 0.26±0.04d respectively [Table 3]. In Cr contaminated soil alone, the application potash increased the bioconcentration factor (BCFr) value to 0.15±0.02a while in co-contaminated soil, the BCF_R was reduced to 0.07±0.02bc [Table 3].

3.4 Effect of potash on Anthracene dissipation and residual Anthracene concentration in soil

The present result showed that the application of potash in single anthracene and Cr-anthracene co-contaminated soil significantly ($p<0.05$) dissipated anthracene by 71% and 78% respectively [Figure 1].The extractable anthracene decreased significantly ($p\leq0.05$) in single anthracene and Cr- anthracene co-contaminated soil after the application of potash to 29mg/kg and 22mg/kg respectively [Figure 2].

Table 3: Translocation and Bio concentration Factor (TF / BCF) of Cr as affected by potash in the remediation of co -contaminated soil using *Glycinemax L*

Treatment	TF	BCFs	BCFr
Soil+G.max	0.29±0.01a	0.0009±0.00a	0.003±0.00a
100mg/kg Cr+G.max	0.25±0.02a	0.02±0.01ba	0.1±0.01a
100mg/kg Cr+G.max+p	0.09±0.04c	0.01±0.01bc	0.15±0.02a
100mg/kg Cr+PAH+G.max	0.58±0.01a	0.04±0.01b	0.07±0.02bc
100mg/kg Cr+PAH+G.max +p	0.26±0.04d	0.02±0.01c	0.06±0.02b

Different letters indicate significant difference (TukeyHSD,p<0.05). p=potash, PAH=100mg/kg anthracene

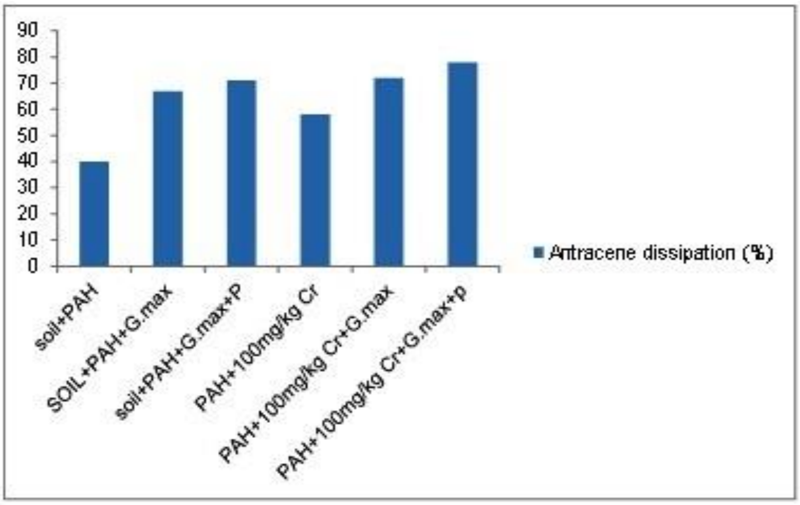

Figure 1: Anthracene Dissipation (%) in planted and non-planted soil as affected by potash in the remediation of co-contaminated soil using *GlycinemaxL*.

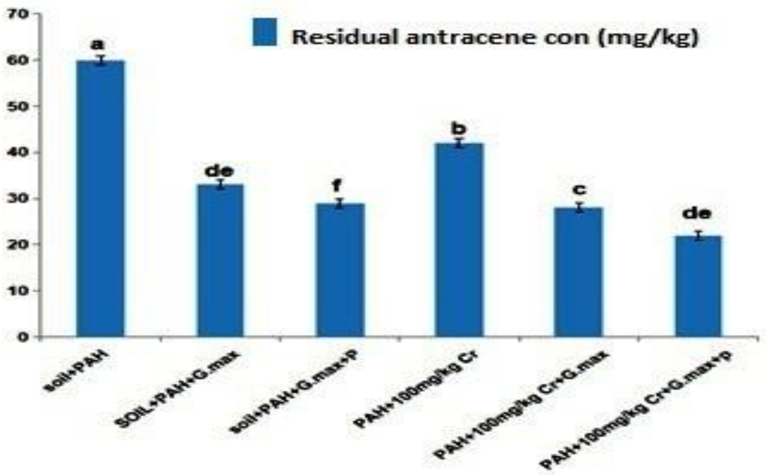

Figure 2: Residual anthracene concentration in planted and non-planted soil as affected by potash in the remediation of co-contaminated soil using *GlycinemaxL*. Different letters indicate significant different (Tukey HSD, P<0.05). n=3, p=potash, PAH=100mg/kg anthrancene.

4.0 DISCUSSION

4.1 Effect of potash on plant biomass and accumulation of Cr

The application potash significantly increased the shoot dry matter weight of *G.max* in soil contaminated with Cr alone and Cr-anthracene co-contaminated soil (Table l).The application of potash reduced the shoot Cr concentration of *G.max* (Table 2).Higher pH values after potash application can result in heavy metal precipitation in soil. This can also reduce the mobility of heavy metal altering their redox state [17]. This mechanism of heavy metal reduction and immobilization enhanced the biomass yield of *G.max* when potash was used as amendment. On the whole, it can be deduced that the presence of appreciable concentrations of anions like nitrate and phosphate, high concentration of potassium in potash justifies its usage as organic fertilizer which enhanced the biomass yield of *G.max* [11, 13]. The application potash significantly increased the shoot dry matter weight of *G.max* in soil contaminated with Cr alone and Cr-antrhacene co-contaminated soil (Table l).

4.2 Residual Antracene in soil

The result of this study show that the dissipation of anthracene in co-contaminated soils and single contaminated soil increased in the presence of amendments. This decrease in the residual anthracene concentration and dissipation after amendment with potash (Figure.1, 2) could also be as a result of the desorption of anthracene which could increase the bioavailable anthracene in contaminated soil. When anthracene is bioavailable, it becomes simple for microorganisms to degrade them. Potash enhanced the dissipation of anthracene with the help of *G.max*, which shows the importance of organic amendments in anthracene dissipation in co-contaminated soils.Also, in this study, the role of planting and non-planting was studied. It was obvious that cultivation of the plant species enhanced the dissipation of anthracene in single and co-contaminated soil [14]. Several processes could have enhanced the dissipation of anthracene in the present study even in non-planted soil. Also because the soil used for the present study was not sterile, it could also be possible that biodegradation by indigenous anthracene degrading microorganisms present in contaminated soil took place.

CONCLUSION

The result of this study show that potash can be applied in single Cr, anthracene and Cr-anthracene co-contaminated soils in the presence of *G.max* for efficient phytoremediation process. Compared to control treatments, the shoot dry weight of *G.max* increased significantly to 0.38±0.0 gpot^{-1} and 0.34±0.02g pot^{-1} with the addition of potash in single Cr contamination and co-contamination with Cr and anthracene. The application of potash in single anthracene and Cr-anthracene co-contaminated soil dissipated anthracene by 71% and 78% respectively. Potash has been proposed as amendments for enhancing the dissipation of anthracene and uptake of Cr in contaminated soil.

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